A CYTOTAXONOMIC STUDY IN VERBESINA (COMPOSITAE)[1]

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In their comprehensive revision of *Verbesina* Robinson and Greenman (1899) recognized 109 species in 12 sections, 4 of which they considered as provisional, unnatural units. During this century Blake (1925) revised section *Lipactinia* and Coleman (1964, 1966a, 1966b) has recently treated 3 others. However since Robinson and Greenman's revision the number of species assigned to *Verbesina* has more than doubled because of descriptions of new species and transfers made into the genus. As a result, the relationships of many species are uncertain and sectional limits have become increasingly obscured. In an attempt to better determine specific relationships and sectional limits, a study of chromosome numbers and crossing relationships is being conducted. This paper represents the initial report on this study.

MATERIALS AND METHODS

The material utilized in this study is listed in Table 1. Voucher specimens are deposited in the herbarium of Indiana University. Crosses were made by rubbing together heads of the individuals being crossed for 2-4 consecutive days to insure that all florets had opened and were pollinated. The crosses were made in insect-free greenhouses at Indiana University with 2-5 plants generally being involved in each series of crosses. Immature heads were fixed in a solution of 95% ethyl alcohol, chloroform and propionic acid (6:2:2) (Jackson 1962). Entire florets were smeared and stained with acetocarmine. Many slides were made permanent with venetian turpentine; selected cells were

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drawn with the aid of a camera lucida. Pollen stainability, taken as a measure of viability, is based on counts of not less than 200 randomly selected grains after having been stained in cotton blue for at least 24 hours. Grains which stained deeply were considered viable; those which stained lightly or not at all were considered inviable.

CHROMOSOME COUNTS

With the addition of the counts presented in this paper, the number of species of *Verbesina* counted is increased to 29, representing about 15% of the genus. Of the 11 species counts here presented, 4 are for previously unreported species. The counts determined for the material included in this study are given in Table 1.

The count of $n = 34$ for *V. alternifolia* and $n = 17$ for *V. occidentalis* are in agreement with previous reports for these species (Heiser & Smith 1955) as are the counts of $n = 17$ for *V. lindenii* and *V. serrata* (Turner, Ellison & King 1961; Turner, Beaman & Rock 1961). Turner and Flyr (1966) report *V. warei* as $n =$ ca 17. A definite report of $n = 17$ is here reported for *V. warei*. The previously unreported species *V. aristata*, *V. longifolia*, *V. rothrockii* and *V. virginica* were determined as having $n = 17$.

Of the species of *Verbesina* thus far reported, 1 has $n = 16$ (Turner, Beaman & Rock 1961), 2 have $n = 18$ (Turner, Ellison & King 1961; DeJong & Longpre 1963) and 3 are tetraploid with $n = 34$ (Heiser & Smith 1955; Coleman 1966b; Turner & Flyr 1966). All other reported species have $n = 17$. A single species, *V. oligocephala*, is reported to have B chromosomes (Coleman 1966a). It would appear very probable that $n = 17$ is in the primitive condition in *Verbesina* and that both aneuploidy and polyploidy have played a role in the evolution of the genus.

CROSSING PROGRAM

The source of the material utilized in the crossing program is listed in Table 1. The crosses made and the results obtained are listed in Table 2. Table 3 presents the pollen stainability and meiotic behavior observed in the hybrids.

Intrasectional crosses: Only 2 sections, *Pterophyton* and

Verbesinaria, are represented by more than a single species in the crossing program. The 3 species of sect. *Pterophyton, V. aristata, V. rothrockii* and *V. warei,* were crossed in every possible combination. The cross *V. warei* × *V. aristata* and its reciprocal both resulted in high seed set; however those of the reciprocal were not grown to maturity. The hybrids displayed intermediacy for several characters. *Verbesina aristata* has an erect habit, rayed heads and numerous disk florets (ca 40-45) whereas *V. warei* is lax-stemmed, rayless and has much fewer disk florets (ca 13-25). The hybrids were intermediate for erectness of stems and possessed rayed heads with numerous disk florets similar to the male parent.

The high pollen stainability of the hybrids as well as the occurrence of 17 bivalents at meiosis supports the morphological evidence that these 2 species are quite closely related. *Verbesina aristata* occurs in southern Georgia and Alabama and in northern Florida, especially in the panhandle region. *Verbesina warei* is endemic to the litoral of the central panhandle region of Florida. Although the ranges of the 2 species are contiguous, ecological isolation is evidently complete and the ranges apparently do not overlap. As would therefore be expected, an examination of herbarium specimens revealed no evidence of natural hybridization.

Intersectional crosses: Nine species from 5 sections were utilized in intersectional crosses. Each of the 4 successful crosses was between species of sects. *Pterophyton* and *Verbesinaria.*

Seeds from the cross *V. aristata (Pterophyton)* × *V. occidentalis (Verbesinaria)* germinated readily to produce robust hybrids. The leaves of the hybrids were intermediate between the petiolate, ovate ones of the male parent and the sessile, oblong ones of the female parent. The wingless stem of *V. aristata* dominated the winged condition found in *V. occidentalis*. The styliferous ray florets of *V. occidentalis* dominated the neutral ones of *V. aristata*. As might be expected for intersectional hybrids, meiosis revealed 34 univalents and pollen stainability was an accordingly low 5%. Although the species are sympatric, no hybrids were detected in the herbarium material examined.

The cross *V. alternifolia (Verbesinaria)* × *V. aristata (Petrophyton)* resulted in a single seed which germinated to produce an initially weak individual. Vegetative morphological characters indicated this plant to be a hybrid. Like the male parent, it possessed wingless stems and the leaves were completely sessile and opposite rather than attenuate-based and alternate as in the female parent. Unfortunately this plant was inadvertently destroyed before material was taken for cytological examination. However pollen stainability was checked and found to be 4%. The low pollen stainability indicates a lack of chromosome pairing and further substantiates the hybrid nature of this individual. The species are sympatric, but no hybrids were encountered in the herbarium material examined.

The hybrids produced by the cross *V. warei (Pterophyton)* × *V. occidentalis (Verbesinaria)* possessed the wingless stems of the female plant and had leaves intermediate between the elliptic-oblong ones of the female parent and the larger, ovate or lanceolate ones of the male parent. The hybrid manifested none of the laxness of stems displayed by *V. warei*. Pollen stainability ranged from 1-4.5% and 34 univalents were observed at meiosis. No indication of natural hybridization was detected in an examination of herbarium material.

Seeds from the cross *V. rothrockii (Pterophyton)* × *V. longifolia (Verbesinaria)* germinated readily to produce both selfs and hybrids. *Verbesina rothrockii* has lanceolate leaves which are mostly opposite below but soon become alternate whereas the linear leaves of *V. longifolia* are frequently subopposite, subwhorled and alternate on the same plant. The hybrids had leaves intermediate in shape which were mostly opposite, the uppermost becoming alternate and possessed linear phyllaries such as are found in *V. longifolia* rather than oblong ones as occur in *V. rothrockii*. Meiosis of the hybrids revealed 17 bivalents with no abnormalities. Pollen stainability was correspondingly high, ranging from 48-60%. Although the species are sympatric in southeastern Arizona, no natural hybrids were detected in the herbarium material examined.

CONCLUSIONS

The crossing program here reported involves too small a portion of the genus to permit broad generalizations. However some conclusions are in order. Although Robinson and Greenman (1899) retained *V. alternifolia* in the genus *Actinomeris*, there has been a tendency in recent years to treat this small genus of 2 species in *Verbesina*. The morphological characters, divergent pappus awns and globose receptacles, by which it is kept apart from *Verbesina* are weak. That an apparent hybrid was obtained between *V. alternifolia* and *V. aristata* further supports the morphological evidence that there is no justification for maintaining the genus *Actinomeris*.

The ease with which hybrids were obtained between sections *Verbesinaria* and *Pterophyton* suggests an affinity on the part of some of the members of these sections. The low pollen stainability and the lack of chromosome pairing in 3 of these successful intersectional crosses does not indicate a particularly close relationship. However the high pollen stainability and the occurrence of 17 bivalents with no abnormalities in the hybrids of the cross *V. rothrockii* × *V. longifolia* indicates a low genetic isolation and suggests the possibility that *V. longifolia* and its relatives may be closely related to some members of sect. *Pterophyton*. This also appears possible on morphological grounds. Robinson and Greenman considered sect. *Verbesinaria* to be unnatural, an opinion with which I concur. Additional crosses may be of aid in understanding the relationships of the species of this section. Since only sects. *Pterophyton* and *Verbesinaria* were represented by more than a single species in the crossing program, the failure to secure hybrids between other sections can not be taken as indicating that genetic isolation is complete between these sections.

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Table 1

MATERIAL USED IN CYTOLOGICAL AND CROSSING STUDIES

Species	Section	$n =$	Source
alternifolia (L.) Britt.	Verbesinaria	34	Bloomington, Monroe Co., Ind. *V6132*
aristata (Ell.) Heller	Pterophyton	17[a]	Lloyd, Jefferson Co., Fla. *V6123*
encelioides (Cav.) Benth. & Hook. f.	Ximenesia	17	Bisbee, Cochise Co., Ariz. *V6219*
encelioides (Cav.) Benth. & Hook. f.	Ximenesia	17	Floresville, Wilson Co., Tex. *V6117*
lindenii (Sch. Bip.) Blake	Saubenetia	17	Isthmus of Tehuantepec, Mexico. *V6138*
longifolia (A. Gray) A. Gray	Verbesinaria	17[a]	Ft. Huachuca, Cochise Co., Ariz. *V6135*
occidentalis (L.) Walt.	Verbesinaria	17	Whitehall, Clark Co., Ga. *V6210*
oligocephala Johnston	Sonoricola	17	Puerto Escondido, Baja California, Mexico. *V6233*
rothrockii Rob. & Greenm.	Pterophyton	17[a]	Mule Mountains, Cochise Co., Ariz. *V6133*
serrata Cav.	Saubenetia	17	Queretaro, Queretaro, Mexico. *144*
virginica L.	Ochractinia	17[a]	Tallahassee, Leon Co., Fla. *V6131*
warei (Nutt.) A. Gray	Pterophyton	17	Wewahitchka, Gulf Co., Fla. *V6130*

[a]Previously unreported counts

Table 2 CROSSING PROGRAM

Crosses	Total Attempts (florets)	% Seed Set	Comments
alternifolia × aristata	174	0.6	probable hybrid
× occidentalis	144	0.0	
× virginica	41	0.0	
× warei	81	0.0	
aristata × alternifolia	165	4.8	apparent self
× occidentalis	169	40.2	hybrids
× rothrockii	199	0.0	
× virginica	109	0.0	
× warei	65	29.2	probable hybrids
encelioides × oligocephala	207	0.0	
× virginica	190	0.0	
longifolia × rothrockii	111	8.8	no germination
occidentalis × alternifolia	101	11.9	selfs
× virginica	309	16.5	probable selfs
oligocephala × encelioides	33	0.0	
× rothrockii	55	47.3	selfs
rothrockii × aristata	90	0.0	
× encelioides	110	0.0	
× longifolia	196	15.6	selfs and hybrids
× occidentalis	77	0.0	
× warei	85	4.7	no germination
Virginica × encelioides	108	0.0	
× occidentalis	492	2.7	probable selfs
× oligocephala	120	0.0	
warei × alternifolia	28	0.0	
× aristata	55	25.5	hybrids
× rothrockii	23	0.0	
× occidentalis	58	12.1	hybrids
× virginica	64	0.0	

Table 3

ANALSIS OF HYBRIDS

Cross	Pollen Stainability %			Meiosis	
	Range	Average	No. Plants	Pairing	No. Plants
alternifolia × aristata	—	4	1	——	0
aristata × occidentalis	—	5	1	34 I	1
rothrockii × longifolia	48-60	53	3	17 II	2
warei × aristata	50-69	62	6	17 II	1
warei × occidentalis	1-5	2	2	34 I	1

LITERATURE CITED

BLAKE, S. F. 1925. On the status of the genus *Chaenocephalis* with a review of the section *Lipactinia* of *Verbesina*. Amer. J. Bot. **12**: 625-640.

COLEMAN, J. R. 1964. A taxonomic revision of sections *Pterophyton*, *Sonoricola* and *Ximenesia* of the genus Verbesina (Compositae). Unpublished Ph.D. thesis. Indiana University.

—————— 1966a. A taxonomic revision of section *Sonoricola* of the genus *Verbesina* (Compositae). Madrono **18** (5): 129-137.

—————— 1966b. A taxonomic revision of section *Ximenesia* of the genus *Verbesina* (Compositae). Amer. Midl. Nat. **76** (2): 475-481.

DEJONG, D. C. D. and E. K. LONGPRE. 1963. Chromosome studies in Mexican Compositae. Rhodora **65**: 225-239.

HEISER, C. B. and D. M. SMITH. 1955. New chromosome numbers in *Helianthus* and related genera (Compositae). Proc. Amer. Acad. Sci. **64**: 250-253.

JACKSON, R. C. 1962. Interspecific hybridization in *Haplopappus* and its bearing on chromosome evolution in the *Blepharodon* section. Amer. J. Bot. **49**: 119-132.

ROBINSON, B. L. and J. M. GREENMAN. 1899. A synopsis of the genus *Verbesina*, with an analytical key to the species. Proc. Amer. Acad. **34**: 534-564.

TURNER, B. L., J. H. BEAMAN and H. F. L. ROCK. 1961. Chromosome numbers in the Compositae. V. Mexican and Guatemalan species. Rhodora **63**: 121-129.

——————, W. L. ELLISON and R. M. KING. 1961. Chromosome numbers in the Compositae. IV. North American species with phyletic interpretation. Amer. J. Bot. **48**: 216-223.

—————— and D. FLYR. 1966. Chromosome numbers in the Compositae. X. North American species. Amer. J. Bot. **53**: 24-33.